THE CONNECTIVE TISSUE CHARACTER OF THE SEPTA OF THE SPINAL CORD AS STUDIED BY A NEW STAIN.*

By Alfred Reginald Allen, M.D.,

OF PHILADELPHIA.

FROM THE DEPARTMENT OF NEUROLOGY AND THE LABORATORY OF NEUROPATHOLOGY OF THE UNIVERSITY OF PENNSYLVANIA.

In working on this new method of staining nervous tissue, I have been able to demonstrate the fact that the posterior medium septum of the spinal cord, as well as a number of the septa of the lateral columns, contain connective tissue which is continuous with the pia. This statement is at absolute variance with the teaching that these septa are composed of exclusively neuroglia tissue.

In staining to demonstrate these pial septa the method is as follows:—

(a) The material should be hardened as if for staining by the Nissl method.

(b) Sections, after washing in distilled water, are placed in a 0.5% aqueous solution of azure II. (or azure I.) and steamed for two or three minutes and then allowed to cool for about fifteen minutes.

(c) Wash in distilled water.

(d) Place in absolute alcohol and gently agitate until clouds of blue cease to be given off.

(e) Place in a mixture composed of carbolic acid 20 toluol (boiling point 110°-112°) 80 for about one minute.

(f) Place in a solution of eosin (Grübler w. g.) in absolute alcohol (alcohol 60, eosin 0.02-0.05), and as soon as the change from dark blue to purple becomes pronounced, remove at once and—

(g) Place in carbol-xylol (25% solution of carbolic acid in xylol). Remove section from this before it turns pink, as after this the differentiation is greatly impaired.

(h) Place in pure xylol.

(i) Mount in xylol balsam or xylol damar.

The only difficulty that will be experienced by those using this stain for the first time will be the steps marked "f" and "g"

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Only practice will teach when to take the section out of the eosin-alcohol solution. I advise those wishing to obtain good results to pass through two sections, leaving the first in the alcohol eosin solution only five seconds and the second section about a minute, and then examining each to determine the faults of the two extremes.

In a properly stained section the pia is a vivid pink and the subpial neuroglia tissue blue. The tigroid substance of the anterior cornual nerve cells is well brought out, and the structure of the dendritic processes is particularly clear. There is a differentiation in the walls of the blood vessels, the intima and media staining blue, the adventitia pink.

When sections are hardened in Müller's fluid, the nerve cells loose their Nissl granules. If the material be first placed in formalin solution 5% to 10% for two or three days, and after that in Müller's fluid, the Nissl granules are preserved, but in this case the differentiation between the pia and the subpial neuroglia tissue is lost. The hardening in Müller's fluid, after the use of formalin, gives a very beautiful picture, in that it brings out the axis-cylinder processes, staining them blue. In this case the medullary sheath stains blue.

A longitudinal section of the spinal cord shows very clearly the collateral branches of the axis cylinders.

This stain does not give any better results with material kept too long in the hardening fluid than can be obtained with any other process.